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Original Research Article

The Antimicrobial Profile of Common Bacterial Isolates in Neonatal Sepsis: A Retrospective Study at a Tertiary Care Hospital

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Abstract:

Introduction: Neonatal septicemia is the leading cause of neonatal mortality and morbidity in India. In India, the primary cause of infant mortality and morbidity is neonatal septicemia. The goal of the current investigation was to identify the common pathogens isolated from the blood of septicemic newborns in the Neonatal Intensive Care Unit (NICU) and their bacteriological profile and pattern of antibiotic sensitivity.

AIM & Objective: Isolation and identification of pathogens responsible for neonatal septicemia from the blood with their antibiotic susceptibility pattern.

Materials and Methods: For a period of one year, from August 2021 to July 2022, blood samples from cases of clinically suspected neonatal septicemia were subjected to aerobic culture, and the isolates obtained were evaluated for antibiotic susceptibility pattern in accordance with CLSI standards.

Results: Four hundred blood samples were examined; of these, 168 (42%) had a positive blood culture. Of these, 50 (29%) were female, 118 (58.4%) were male, and 92 (54.7%) were preterm. Gram negative organisms have been isolated from 66 cases (392 %) and Gram positive organisms from 64 cases (38.09%). The majority of the organisms in this study were gram negative. Of the gram-positive bacteria, 28 (45%) were staphylococcus epidermidis and 10 (0.16%) were staphylococcus aureus. Out of the total number of isolates, 24 (38.7%) were candida, of which 21 (0.33%) were candida auris, 3 were candida albicans, and only 2 (0.03%) were streptococci. One instance of Acinitobacter (0.01%), 25 (37.8%) Klebsiella species, and 29 (43.9%) Gram-negative E. Coli species were all isolated.

Antibiotic susceptibility pattern: Two enterococci (20%) and five gram-negative isolates (0.17%) both exhibited vancomycin resistance. Methicillin resistance was observed in just 2 (0.05%) of the gram-positive Staphylococcus aureus isolates.

Conclusion: This study emphasizes how multi-drug-resistant bacteria have emerged in our setup and how Gram-negative organisms are mostly responsible for newborn sepsis. Considering the changing spectrum of organisms that cause newborn septicemia and their patterns of antibiotic susceptibility, it is important to periodically examine cases in order to identify any evolving trends in the infecting organisms and their antimicrobial susceptibility.

Keywords: Neonatal Sepsis, MRSA, ESBL, NICU.

Introduction

Neonatal septicemia is defined as a clinical syndrome characterized by systemic signs and symptoms of inflammatory response following the appearance or confirmation of infection during the first month of life.

About 25% of neonatal mortality worldwide, primarily in developing nations, is caused by neonatal septicemia. Concern has been raised about the increased prevalence of methicillin-resistant staphylococcus aureus (MRSA), multiple drug-resistant (MDR) strains, and extended spectrum beta-lactamases (ESBLs) in neonatal intensive care units (NICUs) across the globe. [1,2,3] There are two types of septicemia: late-onset septicemia (LOS) and early-onset septicemia (EOS). [4] The most frequently found microorganisms associated with EOS are Haemophilus influenzae, Listeria monocytogene, Escherichia coli, Group B Streptococcus (GBS), and coagulase negative Staphylococcus species (CONS).

On the contrary, LOS is caused by CONS, S. aureus, E. coli, Klebsiella spp., Pseudomonas spp.,

Enterobacter spp., Candida spp., GBS, Serratia spp., Acinetobacter spp., and anaerobes. [5,6]

To reduce the morbidity and mortality of neonates, it is important to choose the right antibiotic therapy based on knowledge of common infections and antimicrobial susceptibility patterns that cause septicemia. Geographically heterogeneous patterns of antibiotic susceptibility are influenced by common antibiotic usage in newborn facilities as well as prevailing local infections.

After clearance from the Institutional Review Board and Institutional Ethics Committee, a cross-sectional study was conducted during August 2021 and July 2022 in the NICU of Anantapur Medical College and General Hospital Anantapur.

Material and Methods

The present study was a cross sectional study. A total of 400 newborns being cared for in the NICU of Anantapur Medical College and general hospital from August 2021 to July 2022. Signs and symptoms such as fever, poor feeding, respiratory distress, cyanosis, cold clammy skin, tachycardia, convulsions, hyperrefexia, jaundice, instability, etc. were taken into consideration at the time of selection.

The inclusion criteria were as follows:

1. Newborns with a clinical diagnosis of septicemia confirmed by blood culture (age≤28 days);
2. Clinical data kept intact.

The exclusion criteria were as follows:

1. Duration of hospital stay <24 hours;
2. Newborns with congenital metabolic diseases;
3. Newborns with septicemia who have been treated with antimicrobials.

Sampling Procedure and Processing: About 1-2 ml of blood was drawn aseptically before starting antimicrobial therapy and directly inoculated into Brain Heart Infusion broth (BHI) (HiMedia, India) in a ratio of blood: BHI of 1:5.The blood culture bottles were immediately sent to the microbiology laboratory and incubated at 37◦ C for 24 hrs and sub cultured on Mac Conkey agar, blood agar, and chocolate agar (HiMedia, India) daily for 7 days.

The inoculated MacConkey agar plates were incubated aerobically, whereas blood agar and chocolate agar plates were incubated in CO2 enriched humid atmosphere using candle jar, at 37◦C for 24-48 hours. Blood culture bottles showing no growth on subculture done after incubation of 7 days were reported as negative. All the

Collected blood samples were processed for culture and isolation by standard microbiological methods. [7]

Antibiotic Susceptibility Testing: The antimicrobial susceptibility testing was done by Kirby-Bauer disc diffusion method as recommended by Clinical Laboratory Standard Institute (CLSI) guidelines. Antibiotic disks (HiMedia, India) used were ampicillin/sulbactam, amikacin, ceftriaxone , cefotaxime, cotrimoxazole, clindamycin, cefoxitin, cefixime, Cloxacillin, erythromycin, gentamicin, meropenem nalidixic acid, ofloxacin, pi peracillin/tazobactam, teicoplanin, and vancomycin. For quality control of antimicrobial susceptibility testing, E. coliATCC 25922 and S. aureusATCC25923 were used. [8]

Ethical Committee Approval: Ethical approval was obtained from the Anantapur Medical College and general hospital -Institutional Review Committee before starting the study.

Statistical Analysis: The collected data were summarized, presented, and analyzed using the software SPSS version 20.

Results

Out of 400 blood samples studied, 168 (42%) were positive blood culture positive. 118 (58.4%) were males and 50 (29%) females and 92 (54.7%) were preterm. Gram positive organisms were isolated in 64(38.09%), of cases and Gram negative organisms were isolated in 66(39.2%), of cases.

In this study gram negative organisms were predominant. Among gram positive bacteria, 10(0.16%) were staphylococcus aureus and 28 (43%) were staphylococcus epidermidis.24 (37%) of them were Candida isolates, among which 21(32%) were candida albicans and 3(4.54%) of them were candida auris and only 2 streptococci (0.03%), were isolated.

A total of 29 (43.9%) Gram negative E.coli species were isolated, and 25 (37.8%) Klebsiella species and one case of Acinitobacter (0.01%) were isolated.

Antibiotic susceptibility pattern: 5 of gram negative isolate (0.17%) showed ESBL production and 2 of enterococci (20%) showed Vancomycin resistance. Among gram positive staphylococcus aureus only 2 (0.05%) isolates showed methicillin resistance.

Table 1: Gram Positive and Gram negative pathogens isolated

Gram Positive	Gram negative
Staphylococcus aureus 10(0.16%)	E.coli 25 (43.9%)
Staphylococcus epidermidis 28 (43%)	Klebsiella 25 (37.8%)
Non albicans candida 21(33%).	Acinitobacter (0.01%)
candida auris 3(0.04%)	

Streptococci 2 (0.03%).	

Table 2: Antimicrobial agents (microgram/disc)

Organism	Amk	Amp	Cefo	Cefta	Oflo	Co-T	Gen	Imp	Car	Pip	Mero
Klebsiella pneumoniae	32% (R)	71% (R)	30% (R)	30% (R)	- S	22% (R)	13% (R)	- S	- S	- S	- S
Esch.coli	20% R	58% R	30% R	30% R	- S	25% R	25% R	- S	- S	- S	- S
Acinetobacter	R	R	R	R	S	S	R	S	S	S	S
Pseudomonas	R	R	R	R	S	R	R	S	S	R	S
Proteus	R	R	R	R	S	-	R	S	S	R	S
Staph. Aureus	30% R	80% R	80% R	60% R	S	50% R	35% R	S	S	18% R	S
CONS	30% R	80% R	70% R	42% S	S	S	28% R	S	S	S	S
Enterococcus	80% R	80% R	69% R	71% R	S	80% R	S(HLG)	S	S	20% R	S
pneumococcus	R	R	R	S	S	S	S	S	S	S	S

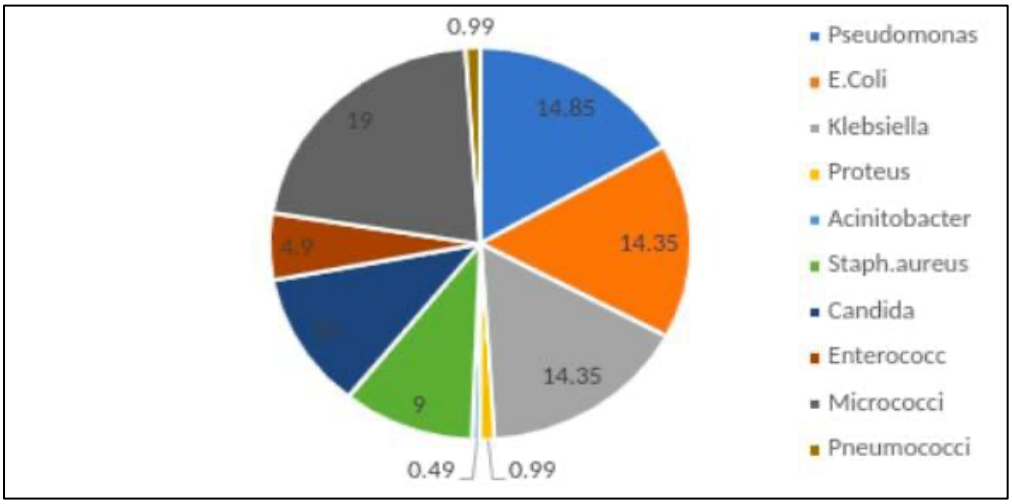

Figure 1: Causative Organisms for Neonatal Septicemia

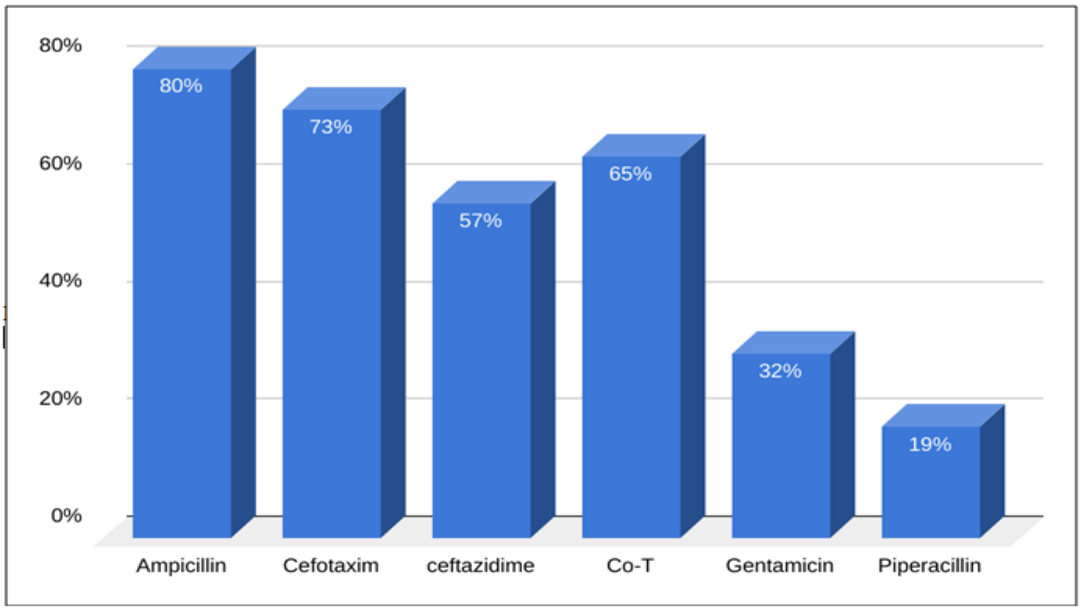

Figure 2: Antibiotic resistance pattern among Gram-positive isolates

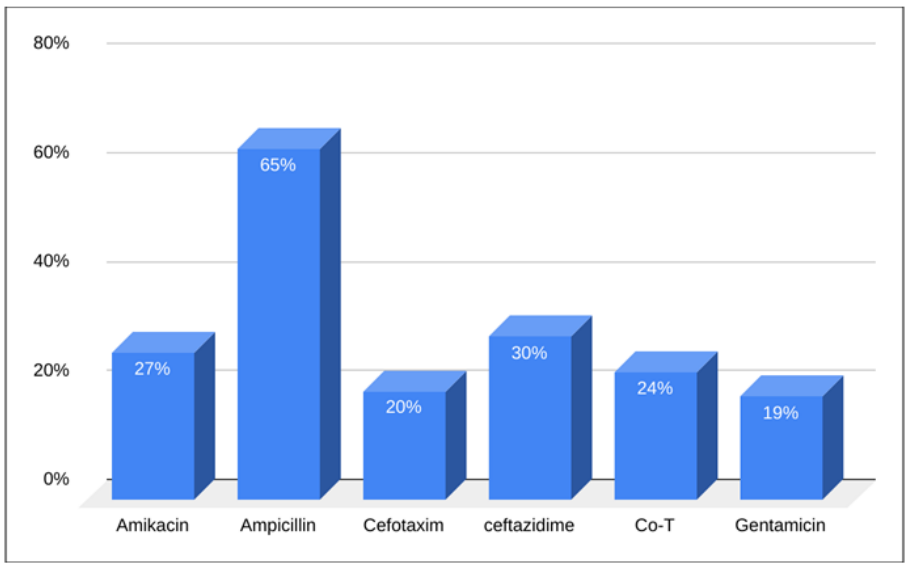

Figure 3: Antibiotic resistance pattern among Gram-negative isolates

Discussion

Neonatal morbidity and mortality are still primarily related to septicemia. To start effective and timely therapy for newborn septicemia, an early diagnosis is crucial. Gram-negative bacteria accounted for 39.2% of the septicemia cases in the current study. A recent study conducted in Karnataka reported 70.5% neonatal septicemia cases caused by Gram-negative isolates.

In the present study E.coli was found to be the predominant pathogen followed by klebsiella accounting for 43.9% and 37.8% cases respectively. Contrary to this study K. pneumoniae was reported as a predominant pathogen in NNPD Report and by Mane et al., Roy et al., and Mustafa et al., from India and by Iregbu et al., from Nigeria. [9,10,11] Other Gram-negative organisms isolated were streptococcus, Acinetobacter species accounts for 0.03% and 0.01%. Acinetobacter species causing septicemia in neonates were reported by Arora et al., and Vinodkumar et al.,

Acinetobacter poses a major problem in NICU. [12,13] After K. pneumoniae, S. aureus was the second most frequent pathogen, isolated from 22.9% of patients. S. aureus as a major pathogen of neonatal septicemia has been reported by Karthikeyan et al14.These findings have implications for therapy and infection control.

K. pneumoniae and S. aureus can survive in the environment for a relatively long time and fairly widely distributed in the hospital environment and therefore have the potential for being transmitted from the environment to the patients through practices that breach infection control measures. [15] Among gram positive bacteria,10 (0.16%) were staphylococcus aureus and 28 (45%) were staphylococcus epidermidis. S. aureus can survive in the environment for a relatively long time and is fairly widely distributed in the hospital environment and therefore have the potential for being transmitted from the environment to the patients through practices that breach infection control measures. In the present study a total of 24(38.7%) Candida isolates were identified, among which 21(0.33%) were candida albicans and 3(0.04%) of them were candida auris and only 2 streptococci (0.03%), were isolated. Predominance of K. Pneumoniae as the causative agent of neonatal sepsis may be due to the selective pressure of antimicrobial agents so that the resistant microorganisms tend to colonize and proliferate in neonates. Some of the S. aureus isolates were found to be methicillin resistant and E. Coli isolates were ESBL producers.

So it would therefore appear that choice of drug for empiric treatment of suspected neonatal septicemia is likely to be difficult in the presence of MRSA and ESBL producers which often failed to achieve therapeutic goals even after showing in vitro susceptibility.

Maximum sensitivity for ciprofloxacin and amikacin has exhibited not only by K. Pneumonia but even the rest of the gram negative and gram positive isolates. This implicates that these two antibiotics can be included as empirical therapy for neonatal septicemia

Conclusion:

This study concludes that gram negative organisms has predominant role in empiric therapy for suspected neonatal septicemia should cover both Gram-negative bacilli and Gram-positive cocci particularly Klebsiella pneumoniae and Staphylococcus aureus. An effective infection control programme, regular antibiotic susceptibility surveillance and evaluation, and the enforcement and periodic review of the antibiotic policy of the

hospital as well as the encouragement of rational antibiotic use will reduce the rates of acquiring nosocomial infections and development of bacterial resistance.

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